ORGANIC RESPONSE[1]

D. T. MACDOUGAL, Ph.D., LL.D.

Desert Laboratory, Tucson, Ariz.

At no time in the history of natural science has such a large share of thought and research energy been directed to the solution of evolutionary problems as at present. Methods of work, plans for experimentation and modes of interpretation have recently undergone such rapid development and improvement that our potentiality for solving questions in heredity and origination is vastly greater than even at such recent date as the beginning of this new century. With increased facility in attack has also come wider vision and altered viewpoints with regard to almost all phases of biology.

Biological thought once quickened and broadened by evolutionary ideas was by this same means led to become entangled in a maze of illusive assumptions as to purpose and plan in organisms from which it is being but slowly freed, to view functions as inevitable reactions, however complex they may be. The variables included in the equations of protoplasmic action are numerous and large, but they do not exceed the undefined principles of osmotic action, surface tension and unknown phases of association and dissociation that are concerned in the interplay of substances in the cell, and upon which

[1] Presidential address, Society of American Naturalists, Ithaca, New York, December 29, 1910.

29589 5

depend the chemico-physical relations of tissue compo-
nents and structures of all kinds.   If physiology escapes
the soporific and deadening influence of the vitalistic con-
ceptions, now appearing in some profusion, it may in turn
furnish the secure means for a long and rapid advance
in genetics, and it may be assumed with some certainty
that the chief superstructures of evolutionary science will
be those securely raised upon a foundation of physiolog-
ically tested facts.

In taking this direction, natural history is not alone;
the briefest comprehensive view of the physical sciences
will show that here also the chief advance lies along the
way of the study of energetics, and that the fundamental
problems are those lying about the mode and means of
transformations of energy.

Recent events in the field of evolution comprehend a
number of movements and accomplishments of extraor-
dinary interest.   The rediscovery of the facts of alter-
native inheritance, the formulation of the concepts of
equivalent, balanced, paired or differential characters, the
results of statistical studies of variability, the analyses
of species of various constitution by pedigree cultures, in
which the value of fertilization from various sources is
carefully measured, the distinction of the biotype or geno-
type as a hereditary entity, the possibilities in the ac-
tion of pure lines within a specific group, the cytological
contributions of fact and forecast upon the physical as-
pects of heredity, and lastly the presentation of the facts
and allowable generalizations identified with the muta-
tion theory, comprise a series of advances, of accretions
to knowledge, furnish a broadened foundation for biolog-
ical science, and disclose additional possibilities in all lines
of experimental research with living things, besides open-
ing up new realms for speculative thought, and stimulat-
ing the scientific imagination to renewed fruitfulness.

Biological literature has also been recently enriched by
a series of formal papers commemorative of the life and
work of Charles Darwin, by more than fourscore workers

representing the laboratories and national cultures of the world. This group of addresses and essays, fortunately written chiefly within four languages, taken collectively, constitutes a critical and evaluatory discussion of the mass of fact and galaxy of theory concerning organic evolution, and furnishes the most complete and thorough appraisal ever made of any subject in modern biology.

The moment, therefore, is one of consciousness of achievement, of realization of increased powers of penetration, and charged with desire for the exploitation of the unknown, and is vibrant with the inspiration coming from such a rapid march of events. With this quickening in activity, the outcries of acrid controversies no longer monopolize our attention, but it must not be supposed that differences of opinion have vanished from among us. The agreement as to the value of methods of experimentation and calibration is a most gratifying fact, but the harmonies of opinion as to interpretation of results have not yet come to a monotone.

On the contrary, the pressure of new and undisciplined evidence has awakened a freshened chorus of voices crying the virtues of special interests and extolling the sufficiency of theories dignified by age and more or less weighty with authority. Those busy with vitalism of various patterns have spun a moiety of their favorite fabric to mend the breaks in the fragile web made by the impact of new facts. Isolation and the mechanism of geographical distribution have again been elaborated to account for all differentiation and what their exponents are pleased to term speciation. The anticipatory formation of structures in a rudimentary condition with a long prefunctional progress, guided by the morphological possibilities and actuated by internal impulses, has again been offered to us, fortified by paleontological fact and clever logic, in such manner as to avoid most of the serious objections to orthogenesis except those of physiological morphology.

Natural selection with diverse meanings and manifold implications has been made to explain development, differentiation and general evolutionary progress. The tumult is greatest at the present time, however, about the idea of mutation. Standing to one side, the biologist hears a medley of assertions "that mutations have long been known," "do not exist," "were discovered by Darwin," "are always an evidence of hybridization," "result in the formation of nothing but elementary species," "give only weakened derivatives that are quickly swamped by parental forms," "are encountered only among cultivated plants," "the mutation theory is based upon the conception of unit characters," "constitutes the only adequate means of accounting for the enormous number of living forms and myriad characters of living things," "unit characters are unreal, have never been seen, do not exist and are incapable of demonstration." "The difference between mutation and variation is one of amplitude only," and lastly mutation signally "refutes Darwinism," and "swings us back in harmony with the theologian's arguments for special creation."

The absurdity of the many injudicial assertions by the partisans concerned need not blind us to the stubborn fact that saltatory changes do occur in hereditary pure lines in a large number of forms in both plants and animals. Observations and experiments have established beyond doubt that mutation is one way by which organisms bearing new combinations of qualities may arise, although it is probable that its importance as a general procedure varies in different groups of organisms and certain that many shades of opinion as to its exact part in the evolution of living things will always be held.

Our appraisement of the value of all the protheses cited may also be amended from time to time with viewpoints altered by the advance of knowledge. The situation with regard to one hypothesis is far more serious, however. This is the theory which predicates direct adaptational adjustment of the organism, quickly or

slowly as the case may be, to environic factors, and
the inheritance of the somatic alterations constituting
such variations. The various corollaries of this theory
have the force of a certain obviousness, its assumptions
have been of ready service to the systematist and bio-
geographer, and its conclusions have long been tolerated
in the absence of decisive tests which are not to be easily
made or readily carried out. The time has now arrived,
however, when the claimants for Neo-Lamarckianism and
all of its conclusions must show cause for its further con-
sideration, or else allow it to drop from the position of
being seriously taken as a method of evolutionary advance.

It is unanimously agreed that organisms, plants as well
as animals, change individually in aspect, in form and
structure of organs, in functionation and habit as they
encounter swamps, saline areas, gravelly uplands or
slopes, climatic differences identifiable with latitude or
elevation, and other physical and biological factors. It is
assumed that these somatic alterations are accommodative
and adaptive, making the organism more suitable for the
conditions which produce the changes. Such an assump-
tion is an over-reaching one. Any analysis of the changes
which an organism undergoes after transportation to a
new habitat will disclose one or a few alterations which
might be of advantage in dealing with the newly encoun-
tered conditions, but with these are many others, direct,
necessitous, atrophic, or hypertrophic as to organs which
have no relation whatever to usefulness or fitness. Fur-
ther, a critical examination fails to disclose any theoret-
ical considerations or any actual facts which would con-
nect inevitably the somatic response with the nature of
the excitation, outside of the specialized tropisms in
which specific reactions are displayed. Even in these
the adjustment is of such nature that a mechanism spe-
cially perceptive to contact, for example, may react to
changes in temperature, as illustrated by the action of
tendrils, and many similar cases might be cited. It is
evident that the soma of a plant or animal is not to be

considered as capable of adaptive alterations to every new agency which may cause changes in its form, structure or functionation.

Next we come to the very crux of the whole matter: do the unusual forms or activities of organs resulting from environic causes act in any manner upon the germ-plasm connected with such altered bodies? If we are to consider the activities of the organism or of the cell to depend mainly upon its chemical structure and constituency and such a generalization seems unavoidable, then we have means by which the soma might cause its properties to be reflected from the germ-plasm in a succeeding generation, since the chemical mechanism of the soma and germ-plasm must be of the most intimate nature. That some such connection does actually exist is strongly suggested by the behavior of a great number of organisms which have been seen to carry marked environic effects to the second or even third generations; if the interrogation be made as to why the induced qualities are carried no further it may be said that the reply may be suggested by the results of long-continued action of the exciting agency, such as has been used by Woltereck with *Daphnia*.

If a general view be taken of the available information of interest in this connection, three classes of facts will be discerned. One group is comprised in the mass of information obtained by the operations of the horticulturist, the agriculturist and the breeder as to the behavior of crops, plants and domestic animals, when transferred from one habitat to another. The greater part of such data is the result of observations which do not comply with the ordinary requirements in the avoidance of error so that strict comparisons as to the behavior of organisms under conditions of various habitats are impossible. A consideration of the literature yields many suggestions for experimental research and the simple generalization that the direct effects of climatic complexes on the seasonal cycle, and upon color, or struc-

tural features of the individual, may be repeated or car-
ried over two or three generations, in a habitat where
the specific causal combinations are lacking. This is the
available total of knowledge furnished us by economic
operations, and by the introduction operations of botan-
ical gardens and plantations.

In contrast with these the fortunate experience of Zed-
erbauer with *Capsella* has yielded some conclusions of
exceptional importance. A genotype of *Capsella Bursa-
pastoris* resembling *taraxicafolium* was found on the
lower plains of Asia Minor, and displayed the well-
known characters of this form, including broad leaves,
whitish flowers, and stems 30–40 cm. high. A highway
leads from these regions to a plateau at an elevation of
2,000 to 2,400 meters. The conditions of distribution are
such as to indicate that the plant has been carried up
this thoroughfare by man, and in this elevated habitat
it has taken on certain alpine characters, including elon-
gated roots, xerophytic leaves, stems 2–5 cm. high, red-
dish flowers, with a noticeable increase of the hairiness
of the entire plant. That the distributional history has
been correctly apprehended seems entirely confirmed by
the fact that when seeds are taken from the lowlands the
alpine characters enumerated are displayed at once as a
direct somatic response. When seeds are taken from
plants on the elevated plateau where their ancestors may
have been for many years or many centuries (perhaps as
long as 2,000 years) and sowed at Vienna and in other cul-
tures carried through four generations the leaves lose
their xerophytic form and structure, but the other charac-
ters are retained within the limits of variability. The
stems show an increase in average length of 1 or 2 cm.,
the roots change as much, but the reproductive branches
and floral organs retain their alpine characters. The
slight modifications undergone by these features were
seen to reach a maximum and to decrease in the latest
generations cultivated. The structural changes and im-
plied functional accommodations are indubitably direct

somatic responses, there is no escape from the conclusion that the impress of the alpine climate on the soma has been communicated to the germ-plasm directly or indirectly in such manner as to be transmissible, and the suggestion lies near that repeated and continued excitation by climatic factors may have been the essential factor in such fixation.[2]

Among the most noteworthy investigations of the features of interest in connection with habitat changes are those being made by the anthropologist in which somatic calibrations of immigrating races and linguistic studies of peoples of known origin, geographical movement, and established relationship are being used to great advantage. No more fascinating chapters of scientific literature are to be found than those which delineate the migratory movements, segregation and habital reactions of Polynesian islanders, of North American Indians, or of Asiatic peoples, yet their value as actual contributions to the phase of biology of interest to this society is hardly recognized. The investigator of problems in anthropology has the advantage of dealing with an animal whose psychology, history, traditions and records are readily intelligible to him, so that a much wider range of facts may be brought within the zone of reliability than when we deal with an organism whose actions, at best, are but imperfectly understood by us.[3]

A second series of results of great interest and suggestiveness are those which have been obtained in various laboratories as to the individual modifications in cyclical activity, functionation and structure of plants and animals in response to unusual stimuli, or under the influence of unusual intensities of the common environic components. The behavior of organisms in constant illumination, equable and variable temperatures, salinity, alkalinity or acidity of the medium, unusual pressures of at-

[2] "Versuche ueber Vererbung erworbener Eigenschaften bei *Capsella bursa pastoris*," *Oester. Bot. Zeitschr.*, Vol. 58: pp. 231–236, 285–288, 1908.

[3] See Boas, F., "Changes in Bodily Form of Descendants of Immigrants," The Immigration Committee, Document No. 208, presented to the 61st Congress, 2d Session, Washington, D. C., U. S. A., 1910.

mospheric constituents, to unusual compounds and unaccustomed food-material, make up an important proportion of the sum total of information ordinarily classified as physiology. The morphogenic and accommodative adjustments presented afford by analysis the best conceptions available as to the nature of the physiologic activity of organisms.

The experimental results of Stockard with fish eggs subjected to the action of various chemical substances are of·unusual interest in the present connection. The cyclopean embryos of *Fundulus* formed in sea-water containing magnesium salts offer the first known example of the induction of an abnormality in the vertebrates occurring in nature, by specific reagents. Suggestion of a common cause is obvious as it is in the instances in which similar divergences have been secured in the laboratory with plants. As will be pointed out later, such analytical tests constitute a very important part of the procedure in the study of acclimatization results.[4]

In very few cases, however, has the permanency or heritability of the deviations induced been tested, and in most of such tests the agencies employed might have acted upon both soma and germ-plasm, as will be apparent upon an examination of the work of Standfuss, Fischer, Pictet and Houssey. The work of these older experimenters has been reviewed so many times that it will be unnecessary to discuss their results further in the present paper. This was done at the Darwin memorial meeting in 1908, and quite recently by Bourne in his address before Section of Zoology of the British Association for the Advancement of Science, at the Sheffield meeting.[5]

The present opportunity may well be used to make a presentation of the results of the last few years obtained

[4] Stockard, C. R., ''The Development of Artificially Produced Fish.— The Cyclopean Embryo,'' *Jour. Exper. Morphology*, Vol. 7, No. 2, p. 285, 1909.

[5] *Nature*, Vol. 84, p. 378, 1910, September 22, 1910.

by investigations, using a more perfected technique, and having the advantage of a keener insight into the real nature of the problems to be solved.

That the general hypothesis with its corollaries is being subjected to the most critical examination and that the assumptions implied in the conception of inheritance of acquired characters are being put to exact and conclusive tests, is readily apparent when a review is made of recent and current researches in which living material from widely separated groups of animals and plants is being subjected to a variety of nutritive conditions and climatic agencies. Klebs, who has long been concerned with the morphogenic reactions of plants, has determined a series of conditions under which the stages of mycelial development, asexual zoospore and sexual or oospore formations in filamentous fungi may be inhibited or variously interchanged. Much more important reactions were obtained from *Sempervivum,* the live-forever of the garden. In this plant, inflorescences were replaced by single flowers by experimental excitation while it was found the number and arrangement of the floral organs as well as of the stamens and carpels could be altered. Furthermore, the deviations in question were found to be transmissible to the second or third generation in guarded seed-reproductions.[6]

Microorganisms with a short cycle offer peculiarly advantageous material by reason of their simple reproductive processes, and also by the fact that it is possible to control environic factors with exactitude. The voluminous literature of bacteriology shows that much attention has been devoted to the building up of characters by selection, and to the study of the behavior of morphological divergences occurring in special cultures.

The experiments of Buchanan with *Streptoccus lacticus* yields the conclusion that phases of fluctuating variations in the bacteria induced by cultures may not be fixed,

[6] Alterations in the development and forms of plants as a result of environment, Proc. Roy. Soc. Lond., Vol. 82, No. B. 559, p. 547, 1910.

and are not transmissible, which is in accord with the main body of evidence upon this point.   There are, however, a number of records of the appearance of definite qualities or morphological characters in the yeasts, which were transmissible and permanent.   These departures were so striking as to be capable of being regarded as mutational, and their origin has been ascribed to the influence of the environment by experimenters of notable skill, such as Beijerinck, Winogradsky, Lepeschkin, Hansen and Barber.   It may be recalled in this connection, that environic responses are generally sudden, and that the entire range of departure may be made in a single generation, at most in two or three.[7]

Pringsheim after a comprehensive review of his own work and of other available evidence obtained by a study of accommodations or adaptations of yeasts and bacteria to unusual temperatures, culture media, and poisons, concludes that some of these variations are fixed and transmissible both asexually and by spores, while others are not.   It is not easy to analyze contributions upon this subject with reference to the differential action of the exciting agencies upon soma or germ-plasm, neither is it clear as to the action of the selection in the experimentation.   It is important, however, to note that the alterations concerned are direct functional responses to the exciting agencies.[8]

The researches of Jennings with paramœcium deals with conditions of morphology and physiology not widely dissimilar from those offered by the bacteria with regard to the present problems, and his work has been carried out with an extensiveness and thoroughness impossible to the worker with more massive and more slowly moving organisms.   Cultures were carried through hundreds of generations with no progressive action in fluctuating

[7] For a brief review of this subject see Buchanan, ''Non-inheritance of impressed variations in *Streptoccus lacticus*,'' *Journal of Infectious Diseases*, Vol. 7, p. 680, 1910.

[8] Pringsheim, H., ''Die Variablität niederer Organismen,'' Berlin, 1910.

variability; while the organism as a whole was strongly resistant to all kinds of environic influences, and actual alterations were extremely rare. Most of the supposedly acquired characters disappeared in two or three generations by fission, although one was followed for twenty-two generations. The new character was borne by only one of the pair produced by a division, except in rare instances, and in only one case was there found such modification as to produce a race bearing the odd character in which the feature in question was imperfectly transmitted in series of asexual generations.[9]

The results of Woltereck with *Daphnia* offer something by way of contrast and also serve to illustrate the necessity for continuation of parallel cultures for the purpose of comparison of divergent forms and the normal. The particular group of this crustacean furnishing the experimental material is taken to be very variable, and it was subjected to over-feeding with the immediate result that the variability of the form of the head appeared to be widened, the size of this structure being increased. This disappeared when lots from the culture were restored to normal conditions in the earlier stage of the work. After three or four months of over-feeding, the form of the head came within narrower limits, and fewer aberrants were seen, while lots returned to normal conditions, showed a slower restoration of the original form of the head. Two years after the cultures were begun, it was found that the original head form was not displayed by young restored to normal nutrition conditions, the larger helmet being persistent. It seems fairly certain that a new genotype resulted from the long-continued action of the culture medium.[10]

[9] Jennings, H. S., ''Heredity and Variation in the Simplest Organisms,'' Amer. Nat., Vol. 43, No. 510, June, 1909; and other papers by the same author.

[10] ''Weitere experimentelle Untersuchungen ueber Artenveranderung speziell ueber das Wesen quantitativer Artenunterschiede bei Daphniden,'' Sonderabdruck a. d. Verhandl. d. Deut. Zool. Gesell., 1909.

In the experiments of Sumner mice reared in a warm room were found to differ considerably from those reared in a cold room in the mean length of the tail, foot, and ear, and the differences were transmitted to the next generation. The differences may be reasonably designated as being directly individual and somatic, and as having been transmitted by the germ-plasm, which was not subject to the action of various temperatures in the first instance. The reaction forms have an additional claim upon our attention, since they are the ones which distinguish northern and southern races of many animals. The crucial test of the value of the alterations induced in the mice is the one applicable to all of the experimentation on this subject, a test in which two parallel series of cultures, one under the altered environment and the other under usual conditions, should be kept going continuously for a long number of years, lots being withdrawn from both, from time to time, for long-continued comparative culture in normal habitat and under other conditions. Effects due solely to fluctuating variability may be expected to reach a maximum and minimum within two or three years, leaving the enduring effects standing alone, or in such relief as to be capable of ready calibration.[11]

Kammerer carried out some tests with salamanders three years ago which have the interest attached to any attempt to interpret geographic or habitat relations. *Salamandra maculosa* is viviparous when it lives high in the mountains and ovo-viparous at lower levels. *S. atra* is an alpine form and the larvae are large with very long gills. When the latter form was kept at unusually high temperatures the larvæ produced resembled those of *S. maculosa* in its lower warmer habitats. *S. maculosa* kept in low temperatures and without water showed a cumulation of effects by which the characters of the

[11] Sumner, F. B., ''An Experimental Study of Somatic Modifications and their Reappearance in the Offspring,'' *Archiv. f. Entwickelungsmechanik d. Organismen*, Vol. 30, pt. 2, p. 317, 1910.

young and the reproductive habits resembled those of
S. atra. The conditions of these experiments are not
such as to allow a definite separation of somatic and ger-
minal effects, neither was the permanency of the newly
acquired habits tested to such an extent as to determine
their hereditary value. That characters and habits may
be modified in such manner as to appear in the next gen-
eration or two in the absence of exciting conditions is il-
lustrated by hundreds of authentic examples in plants
which have long been known.[12]

My own earlier work with relation to this subject con-
sisted chiefly of ovarial treatments in which the main
and accessory reproductive elements of seed-plants were
subjected to the direct action of solutions of various
kinds. New combinations of characters constituting a
distinct elementary species or genotype were obtained in
one plant, and the divergent type has been found to trans-
mit its qualities in the fullest degree as far as tested, to
the fifth generation. Still more marked forms were ob-
tained in a second genus, the divergent progeny being
lost in transference to the Desert Laboratory, while
marked responses have been obtained in the extensions
of these experiments upon species representing widely
different morphological types in Arizona. The greater
majority of the tests have been made upon plants grow-
ing under natural conditions, so that environmental reac-
tion in addition to that of the specific reagents might be
excluded. Progenies representing many species, in-
cluding thousands of individuals, many of which are di-
vergent, are now under observation. Absolute finality
of decision with respect to the standing of the new types
may be reached but slowly.

Gager produced chromosomic aberrations in the reduc-
ing divisions of *Œnothera* by irradiations and such excita-
tion was also followed by the appearance of aberrants in
the progeny, the hereditary qualities of which have not

[12] *Arch. f. Entwickel.*, Vol. 30, pp. 7–51, 1907.

been tested. Using similar excitation Morgan induced
the appearance of white eyes and of short wings in a pedi-
gree culture of the fly, *Drosophila ampelophila.* Both
qualities were sex-limited and mendelized when paired
with the red eyes and long wings of the original type.
Both however seem to be fully transmissible.[13]

A related phase of the subject is that of the interposi-
tion of environic factors in mutations and hybridizations.
DeVries has repeatedly called attention to the fact that
the composition of hybrid progenies of mutants with
each other and with the parental form might be altered
by nutritive conditions, and the author has cited the fact
that mutations were made by *Œnothera Lamarckiana*
in the climate of New York which had never been seen
in Amsterdam. Furthermore, in discussing the diver-
gent results of DeVries and myself, obtained by crossing
the same forms in Amsterdam and New York, the sug-
gestion was made that "the manner in which the var-
ious qualities in the two parents are grouped in the prog-
eny might be capable of a wide range of variation.
Many indications lead to the suggestion that the domi-
nancy and prevalency, latency and recessivity of any
character may be more or less influenced by the condi-
tions attendant upon the hybridization; the operative
factors might include individual qualities as well as ex-
ternal conditions."[14]

Using abnormal temperatures for excitation, Kammerer
induced color changes in *Lacerta* constituting female di-
morphism in one species, and male dimorphism in another,
and the newly induced characters, like the original ones,
behave in a mendelian manner in crosses, although the
heredity has not been carried through enough generations
to test their permanence.[15]

[13] Morgan, T. H., The method of inheritance of two sex-limited charac-
ters in the same animal, Proc. Soc. for Exper. Biol. and Med., Vol. 8, No.
1, p. 17, 1910.

[14] MacDougal, Vail, Shull and Small, ''Mutants and Hybrids of the
Œnotheras,'' Pub. No. 24, p. 57, Carnegie Inst. of Washington, 1905.

[15] Vererbung erzwungener Farbenaenderungen. Arch. f. Entwickl., Vol.
39, Hefte 3 and 4, p. 456, 1910.

Much more striking evidence upon the matter has been recently obtained by Tower in intercrossing *Leptinotarsa decemlineata, L. multitineata, L. oblongata* and other species in their habitats in southern Mexico, and at the desert laboratory. Among other divergences one of the three first generation intermediates characteristic of these cultures was lacking from the Tucson cultures, although two other such forms were included.[16] In a

Fig. 1.   Acclimatization shelters and beetle cages, Desert Laboratory (2,600 ft.).

comprehensive treatment of the entire subject with especial reference to modifications in dominance Tower says:

The experiments and observations herein given warrant the general statement that conditions external to a cross are important factors in determining the results thereof. This conclusion has been worked out in both normal and hybrid crosses, in crosses between races which have been created selectively, and between forms which arose as sports;

[16] See Report, Department of Botanical Research, Carnegie Institution of Washington, for 1908 and 1909.

and the second series of experiments in synthesis is sufficient warrant
for attributing to this factor a considerable importance in evolution.[17]

   Tennent arranged a series of hybridizations of Echino-
derms at Tortugas which yielded data of great interest in
connection with the earlier conclusions of Vernon, Don-
caster, and Herbst as to the influence of temperature and
season changes upon dominance. From the information
derived from crosses of Hipponoë and Toxopneustes it is
clear that the dominance of the parental characters is
dependent upon the alkalinity or the concentration of the
OH ions. The products of the trial cross fertilization,
however, were not reared to maturity.[18]
   No phase of the subject under discussion is more
readily amenable to experimental investigation, and no
results may be expected to bear more directly on the
mechanism of inheritance than those in which similar
unions give dissimilar progenies under the pressure of
unlike environments. It is to be noted that everything
of value with regard to the influence of environment
upon hybridizations has been secured by the introduc-
tion of the geographic or climatic element, that is, the
unions leading to divergent results have been made in
habitats in which the environic complexes differed not in
one, but in many features. Thus the climatic components
in southern Mexico reach dissimilar maxima and minima
and run unlike courses from those of Arizona.
   This method of transplantation of organisms to dis-
tant localities furnishing congeries of climatic factors
markedly different from those of the habitat in which
they were found is one which offers opportunities of the
broadest kind, and such exchanges have been made be-
tween fresh and salt water, cave and surface, alpine

   [17] Tower, W. L., ''The Determination of Dominance and the Modifica-
tion of Behavior in Alternative (Mendelian) Inheritance, by Conditions
Surrounding or Incident upon the Germ Cells at Fertilization. (Reprinted
from *Biological Bulletin*, Vol. XVIII, No. 6, May, 1910.)
   [18] See Report, Director Dept. of Marine Biology, Carnegie Institution of
Washington for 1909 and 1910.

summits and lowland plains, high and low latitudes, with results of somewhat limited value until recently. The first of these in which plants were used was made by Nägeli, who carried on observational work on a large number of species in plantations of the botanical garden at Munich, detecting certain obvious alterations which did not appear to offer anything of hereditary value.

The more recent work of Bonnier was directed chiefly toward comparison of the vegetative activity, anatomical modification, and developmental habit of plants exchanged between the mountain and low-land. The care used in attempting to transport soils with the plant was almost wholly without direct application, since the character of the soil is so largely a function of climate that the course of a single season would suffice to change or materially modify any transported soil. Such a precaution might have the sole merit of furnishing the transplanted species with a limited amount of some compound necessary for its growth, but any small amount of soil becomes quickly permeated by solutions from the formations below and contiguous to it. Bonnier's results include much that is suggestive, although no effects were secured which did not disappear within two or three seasons after a plant had been removed 'from the influence of the exciting agencies or returned to its original habitat.

The first realization of results of importance from cultures widely extended geographically has been obtained in the experiments with Leptinotarsæ by Tower, in which various species of these beetles were studied in their habitats in southern Mexico, in open air and glass houses as far north as Chicago, as far east as the Atlantic and as far west as the Desert Laboratory. Facilities for work upon special problems are now being organized at several places and many contributions to the subject may be expected within the next decade.

The plan for work upon the problems of special interest in connection with the Department of Botanical

Research of the Carnegie Institution of Washington, implies the establishment of experimental cultures in localities which furnish distinct types of climate, or which have characteristic complexes of meteoric factors, as indicated by the vegetation indigenous to them. Secondly, these localities have been chosen with regard to their geographical relations so far as possible, in order that the possible and probable fate of migrating species might be studied. The behavior of plants in these localities is

FIG. 2. Santa Catalina mountains as seen from Desert Laboratory. Experimental plantations shown in figures 3 and 4 are located on this range.

recorded as to anatomical alteration and physiological departure. Having detected some such feature of apparent importance, its reappearance in plants from seeds carried to the original habitat and other locations is followed as one line of evaluation. Contemporaneously, the form is taken into the laboratory and here by analytical experimental tests the effort is made to ascertain to what special agencies the departures are due. Four

main locations furnish the chief facilities for these some-
what extensive experiments, which may be briefly charac-
terized as follows. The domain of the Desert Laboratory
has a subtropical arid climate, with one cool moist sea-
son, one warm wet season, two intervening dry seasons,
the vegetation being chiefly composed of spinose xero-
phytic shrubs and woody plants, with a large number of
the more advanced types of desert plants, which carry an
immense water balance, such as the cacti and other suc-
culents. The total rainfall is 12 inches, relative humid-
ity falls as low as 5 per cent. for extended periods and
the soil moisture remains below 10 per cent. for weeks,
and the altitude is 2,300 feet; maximum temperatures of
112°–114°, minima of about 16° F., with a total exposure
below the freezing point of from 12 to 80 hours per
annum are encountered.

The xero-montane plantation lies at 5,400 feet on the
near-by slopes of the Santa Catalina Mountains at the
extreme upper edge of the characteristic desert flora in
the oak belt of vegetation with a rainfall of 16 to 18
inches per year, minima a few degrees lower than those
of the Desert Laboratory, with such an extension of cold
nights as to make temperature a distinct limiting factor;
relative humidity is extremely low, soil moisture quite as
low as that of the base plantation, and the activity of
vegetation of the winter wet season which is such a
marked feature of the lower plantation is entirely lack-
ing. The meteoric and other agencies carry a constant
stream of seeds from this locality into the region of the
laboratory.

The montane plantation lies at an elevation of 8,000
feet in a forest of pine, spruce and aspen, with a climate
equivalent to that of northern Michigan, the growing
season being about 110 days, the winter being character-
ized by a heavy snowfall and temperatures as much as
20°–25° below zero Fahrenheit. The spring and autumn
are dry, and midsummer has the usual manifestation of
heavy thunder-storms, in which the precipitation is

slightly less than the amount in the winter. The yearly
total is between 35 and 40 inches. The vegetation is
characterized by conifers, grasses and a wide variety of
herbaceous and shrubby perennials, very few annuals
being found here. The mountain streams carry the seeds
of the contiguous elevated slopes and valleys in great
profusion to the region of the xero-montane plantations
and to the lowlands of the character of those around the
Desert Laboratory. These three localities form a con-
nected series in which the behavior of the tested species
may be expected to offer phenomena of wide significance
and of direct bearing on many phases of geographical
distribution and evolutionary advance.

The fourth plantation is at Carmel, California, some
800 miles distant in a straight line from the first three,
within a thousand yards of the Pacific Ocean in a forest
of Monterey pine, the soil being granitic sand, with or-
ganic material or humus in some places, and a heavy
cement in others. The climate is characterized by a win-
ter wet season, in which the minima are scarcely below
the freezing point and the exposure to such low tempera-
tures is for not more than fifteen or twenty hours per
year. A period of heavy continued fogs during two
months of the midsummer results in minima of 41° F.
in July and August, there being almost no precipitation
between March and November. The total precipitation is
about 18 inches per annum. The place, therefore, has
one rainy season, a dry spring and fall, and a cool mid-
summer, conditions exceptionally favorable for the sur-
vival of species introduced from the localities of the
other three plantations of the series. It is obvious that
if the data concerning the climatic factors are integrated
or summarized and placed in parallel columns a ready
means is afforded for detecting the causes which prevent
survival or facilitate the development of any form in
any locality, and a proper analysis of the same facts
may also yield direct suggestions as to the nature of the
excitation responsible for any departure on the part of
a plant removed from one habitat to another.

The groups of species interchanged among the four different localities include material upon which such analysis may be most readily made. In addition, the introductions are also arranged to simulate certain geographical movements and topographical effects. Species from eastern America and from the lower plantations are taken to the montane and xero-montane plantations to meet conditions similar to those they might encounter in a migration toward alpine or arctic regions. Species from the montane locations and from the eastern states are carried to the desert plantations to have the experience of a southward movement, or that of descending mountain valleys, while all of these localities have furnished forms for establishment in the maritime locality characterized by equable conditions in which species may range widely as to latitude and indefinitely as to longitude. The preliminary exchanges included over a hundred species, mostly biennials and perennials; the survivals amount to less than 80, while perhaps not more than a score of these may be expected to yield results of value or interest.

Our increased insight into the nature of natural groups of organisms has shown the necessity and suggested the means of observing certain distinctions and precautions in this work. Thus it is of the greatest importance that the living material shall be shown to be either simple genotypes or that its phaenotypic nature be apprehended in order that the integration and combination of these forms shall not be mistaken for environic effects. When a lot of plants is taken from one plantation to another, data of the original locality are preserved as the stand of the plant in that place serves as the control. If the plant is multiplied vegetatively in the test, it might reveal a possible complex character of the material in bud-sports, but other divergences might be well ascribed to local effects. On the other hand, if introduced in the form of seeds, the possible complex character of the material would soon become apparent, especially if the generations were followed properly. In the actual management of

the cultures, it is found profitable to re-introduce forms
from the original or control lot of various species in
order to follow the first stages of their adjustment re-
peatedly.

The earlier introductions were made in May, 1906, but
the establishment of the system was not completed until
early in 1909. Some of the species have therefore been
observed through the fourth growing season in newly
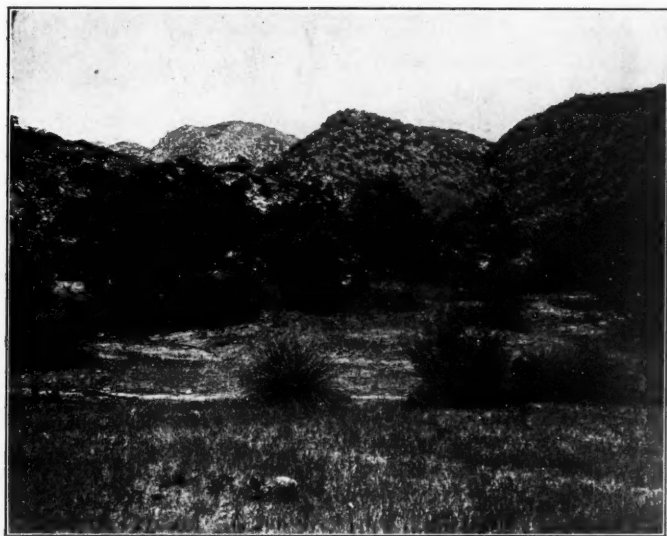encountered habitats, and as the somatic responses are

Fig. 3.   Xero-montane plantation (5,500 ft.), Santa Catalina Mts., Arizona.

immediate or nearly so it may be assumed with some con-
fidence that the alterations observed are those which are
to be tested as to their transmissibility. It may be of
interest to note briefly some of the more salient alterations
with some attention to their geographical significance,
reserving the discussion of structural details for a more
suitable occasion.

1. Many species of perennials native to regions in
eastern America with a temperate summer or growing

season 160–170 days in length, and a winter with extended exposures below the freezing point, endure the climate of the montane plantation with lower minima, shorter growing seasons and more arid dry seasons, all seasonal changes being much more sudden and pronounced than those encountered in their original habitats. *Trillium, Arisæma, Roripa, Sanguisorba, Fragaria* and others offer examples of such survival.

2. Perennials as above survive and thrive in the equable climate of the maritime plantation in which much more equable conditions are found than in the original habitats—*Podophyllum, Sanguisorba, Arisæma, Œnothera, Roripa* and *Fragaria*.

It is to be noted that many species of annuals and perennials are supposed to range from the temperate lowlands of New York to similar montane climates farther south in the Rocky Mountain region, and also to the Pacific coastal belt. Critical examination of material representing the supposed inclusion of the species generally reveals differences denoting elementary species or genotypes which might be grouped together in a Linnean treatment. These relationships offer some most interesting probabilities as to derivation and dissemination which may not be touched upon here.

3. Species from the montane plantation survive and show a luxuriant growth in the maritime plantation with various vegetative modifications, of which *Fragaria, Œnothera, Juglans, Scrophularia, Senecio* and *Dugaldea* offer illustrations.

4. Species from the montane plantation survive and show a development somewhat atypic when carried to the foot of the mountain on which they are indigenous. Examples are *Œnothera, Juglans, Scrophularia* and *Fragaria*.

5. Species from the arid region about the Desert Laboratory survive and show atypic activity in the equable maritime climate. Illustrations are offered by the *Opuntias, Parkinsonia* and *Penstemon*. Species from the

equable maritime location do not survive when taken to
any of the other plantations, with the single exception of
Fragaria Californica, the extremes of temperatures
being the evident limiting factor.[19]

By the consideration of the responses of plants in the
various climates into which they may be introduced in
these experiments, it is possible to determine with some
accuracy the limiting factors acting for the exclusion of
the form in question.   The analysis of the responses to
changed environment may be briefly given as follows:

Species from locations with well-marked seasons, in
which there is a definite contrast between the warm and
dry periods or between dry and rainy seasons, show a
lessened tendency to sexual reproduction, and a conse-
quent weakened capacity to form fruits and seeds when
taken to locations with equable or monotonous conditions.
This is a fact well known to the grower of economic
plants, the chief examples being offered by bush and tree
fruits disseminated to the southward.   Fertilization and
the preliminary stages of seed formation may ensue as
usual, but the absence of the stimulating effect of chang-
ing temperatures usually characterizing the close of a
season appears to be followed by a lack of development
of the fruit.   Examples of this are offered by *Arisæma,
Salomona, Sanguisorba, Actæa, Podophyllum, Menisper-
mum, Apios Fragaria* and *Phytolacca.*   Exceptions are
offered by *Senecio, Œnothera* (some species), *Potentilla,
Geum* and others.

The transplantation of a species from one type of cli-
matic complex to another generally alters the shoot-habit,
or pattern of development of buds.   The maritime loca-
tion is characterized by a profuse development of run-
ners and offsets, and the growth of branches on the lower
part of main axis, above or below ground.   In some spe-
cies, the main axis remains in a very rudimentary condi-
tion.   Excellent illustrations are furnished by *Œnothera,
Scrophularia, Dugaldea* and *Phytolacca.*

[19] See Kuckuck, P., "Ueber die Eingewöhnung von Pflanzen wärmerer
Zonen auf Helgoland," *Bot. Ztg.*, Vol. 68: 49–86, April, 1910.

The removal of the higher types of plants from the desert conditions with which they articulate, that is, extreme forms with reduced shoots and swollen stems, is followed by increased development of spines when grown

FIG. 4.   Montane plantation (8,000 ft.), Santa Catalina Mts., Arizona.

under equable conditions, or in climates with greater water supply as illustrated by *Opuntia santa rita* and other "spineless" forms.

The removal of plants from localities with well-marked seasons to equable maritime climates is followed by a leaf development which may result in the multiplication of the parts. *Fragaria.* All introductions in which the range of climatic conditions to which the plant was subject was narrowed, were followed by increased vegetative activity, which multiplied underground branches and propagative bodies.

The concurrence of these responses in a single form may be well demonstrated by the results of studies of a genotype near *Scrophularia leporella,* found in the vicinity of the montane plantation, which has survived in the shade at the Desert Laboratory and at the seaside locality. In its native habitat, it shows a strict, scarcely branching shoot with a few fleshy succulent roots, which apparently carry water with a small dissolved content. When this form is taken to the Desert Laboratory, its reproductive season is lengthened from two months to five or six months, although but few seeds are formed, the shoots branch more profusely, and a greater mass of underground members are formed. In the maritime location these features are accentuated and the development of branches goes on to such extent that the shoot gives rise to a number of main branches which can not be supplied with water, and hence soon wilt and die. The underground system now includes dozens of thickened members from one to two centimeters in diameter, which may show a total weight of from 6 to 8 kilograms.

The removals of forms included in the experimental series may be taken as fairly parallel to the distributional movements effected by various agencies without the intervention of man. Some, as a matter of fact, are exact duplicates of occurrences in which these same species participate. The alterations noted are undoubtedly environic effects, and may be attributed chiefly to climatic factors. Two common assumptions as to the behavior of plants are to be noted when species are removed to localities widely separated from the habitat in which they are

found. If they fail to survive or do not flourish in the second location, they are said to have failed to adapt themselves to the new conditions. Into this statement may be read one more in accordance with a physiological consideration of the matter to the effect that the intensities of some of the factors present exceeded the maxima of the plants in question and thus acted as limiting factors to their proper or full development or survival.

The second assumption is to the effect that the alterations displayed by a plant in newly encountered habitats are adaptive and that these changes render the organism displaying them better fitted to meet the conditions. Some reactions are of such a nature as to be of benefit to the plant displaying them, but the worker who assumes that this is true of all changes even in species which thrive and luxuriate in the new habitats will soon find himself widely afield from facts capable of being substantiated by experiments. Thus in the case of the *Scrophularia* noted above, the new maritime habitat includes a congerie of agencies which incite it to form enormous clusters of thickened roots and to exhibit the habit of branching densely. So many branches are formed in fact that the conducting channels at the base of the shoot are incapable of carrying a supply of water adequate to the transpiratory needs of the foliar organs, although the vastly increased balance in the root-system would be sufficient to meet the needs of the plant for days, and consequently the widely spreading shoots of these plants show a large proportion of branches which have about reached maturity and are dying. The behavior of the semi-spineless opuntia (*O. santa rita*) offers illustration of the same sort. Bearing only a few or no spines in its native mountains, the new segments in the cool foggy climate of Carmel are spinose at almost every areole. Here the result is very plainly one of the awakening of a latency, since it seems fairly clear that this plant and all of its relatives show spines as a final stage in the reduction of the shoot system, and that the spineless form is the culmination of a line of progress. The

reappearance of the spines is, therefore, one of regression; in a paper before this society a year ago I was able to present results of experimental parasitism, in which the reactions of autophytic green plants when grown as parasites included a number of phenomena, which were not only not adaptive in any sense, but which might reasonably be considered as distinctly unsuitable. Among these was included the very striking autonomic movements of etiolated segments of the prickly pear (*Opuntia*) when it was led to fasten upon other plants as a parasite.[20]

Many alterations in plants in the cultures, however, particularly those concerning the reproductive habits, may readily be interpreted as being adjustments of a directly adaptive character. With these are many correlative changes which are simply carried along. It seems fairly certain that the distinction between the primary adjustive alterations and correlative effects will be made clearer in any analyses made of the possibilities of inheritance of somatic changes. In connection with the discussion of the nature of the parasitic adjustments the behavior of a drop of water when resting upon a rough surface was offered as an illustration of the modifications of an organism under environmental influences. The sectors of the drop in direct contact with a hard object which is not wetted will be most markedly and directly altered, in a manner parallel to the reactions in functions most directly affected by environment, while the free sectors or qualities of the drop or of the organism will be altered in various degrees by correlation stresses.

So far as the responses in the cultures at the four plantations are concerned, they appear to the fullest extent at once and in the first generation. Whether any of them may become fixed and transmissible in a long series of generations subjected to the same conditions, like *Daphnia*, remains to be determined. That this might be the most

[20] MacDougal and Cannon, "The Conditions of Parasitism in Plants," Pub. No. 29, Carnegie Institution of Washington, 1910, p. 37.

important feature of all experiments of this kind was
pointed out three years ago.[21]   Although our attention
has been focused chiefly on the possibilities of the trans-
mission of somatic effects by seed reproduction, yet it is
to be recalled that the continuation of an alteration by
fissions, division or cuttings might come to have great
biological significance.

FIG. 5.   Maritime plantation near the sea-shore, Carurel, California.

Jennings would consider the *Paramœcia* as free germ
cells subject to the direct action of environment, and
themselves propagating by simple division if his meaning
is properly apprehended.   If this is allowable, the same
conception may be extended to include cuttings and all
fission methods of reproduction in plants, even of the
most advanced types.   As a general rule, when a portion
of the sporophyte of a plant, such as an offset, runner,
stolon, tuber, bulb, corm or other detached branch pro-
duces a new individual, the mature characters of the par-
ent disappear in the regeneration or sprouting and the

[21] "Fifty Years of Darwinism," 1909.

ontogenetic procedure of the plantlet will be much like that of a seedling.

The exact observation of the manner in which environic effects may pass the regeneration stage and reappear has not yet been made to any great extent. Doubtless many conditions will be found to affect the process. Bud sports, or vegetative mutations, are, of course, fully transmissible along a series of stages of division by cuttings, and many of them have been found to transmit their divergent characters by seed resulting from close pollination. Mechanically considered, the vegetative reproduction of a plant consists simply of its perpetuation through an unbroken chain of metameres or internodes, each joint arising from a growing point borne terminally or laterally by its predecessor. The projection of induced characters formed by metamere *A* into metamere *B,* therefore, involves the question of germ-plasm as represented by the embryonic mass of the growing points with no opportunity for carrying over structures mechanically as in the *Paramœcium.* The comparative action in heredity when plants are transported to new climates through bulbs and tubers and through seeds is one that has not yet been made, although doubtless horticultural and agricultural literature is rich in the records of facts upon which decisive generalizations might be made.

The genetic character of environic effects remains to be considered. In any species or genotype there is, withal, a limited number of things included within the morphological possibilities. The appearance of any character in an acclimatization culture raises a question at once as to the standing of the new feature. Is it a regressive character, once displayed by the species and now recalled by the very conditions under which it was first induced, or is it to be considered as a character *de novo,* arising simply and directly in response to the external agencies which have been seen to induce it? Thus in the results cited above, our general knowledge of the Cactaceæ leads us to assert with some confidence that the reappearance of a full complement of spines in a prickly

pear from which they had all but disappeared is a regression or return to the condition of the greater majority of the group, a condition which must have been shared by its ancestors at no remote stage in its progressive development.

None of the attempts hitherto made to perfect a theoretical conception which would be useful in interpreting the mechanism of environic responses have had anything more than the most limited usefulness. The stimuli of climatic and many other agencies do not imply the introduction of any strange or new substances into the bodies of the organs affected. These agencies might change the dissociations in such a manner as to modify the relative number of free ions and thus alter the molecular complex of the living matter in a very important manner. The intricate play of enzymatic action might also be altered, and any modification of the relative reaction velocities of the more important processes might result in material and permanent change, especially in those cases in which external agencies interfere directly with the action of the germ-plasm.

The introduction of solutions into ovaries or the exposure of reproductive elements to unusual irradiation may raise the additional liability of disturbed polarity and of modified surface tensions in the cells. It is conceivable that the rearrangement or disturbance of the localizations of substances, especially the mineral salts, might seriously modify the capacities of the bearers of heredity. These direct and material possibilities offer an adequate basis for the organization of experimental research upon the main subject, as well as the means of interpretation of results without recourse to schemes of particulate inheritance or theories as to the constitution of germ-plasm to which may be ascribed usefulness in the discussion of other problems in evolution.

The theoretical consideration of the subject which seeks to assign all cases of inheritance of environic effects to the direct action of the existing agency upon the germ-plasm in itself is one to be regarded with some wariness, as it may lead us into empiric assumptions which may

conceal rather than visualize the actual occurrences. Direct germinal effects are undoubtedly secured in ovarial treatments, and Tower's analytical cultures showed that certain somatic characters induced directly might be secured also by direct excitation of the egg. Such concurrence of reaction may be expected especially with regard to some qualities of simpler organization. Not so readily interpreted are the responses of *Sempervivum*. Alterations in size, number, and structure of floral organs brought about by excitation during ontogeny are surely not coordinate with changes in the germ plasm induced simultaneously. In the case of *Capsella* the transference to an alpine habitat of the plant in the shape of seeds is followed by immediate and direct ontogenetic alterations affecting a multitude of characters. Not until these somatic responses have been repeated, dozens, scores, or perhaps hundreds of times, is an impression made on the germ-plasm that allows it to carry the new characters in the absence of the inducing. These facts suggest to us that the soma is in the closest association with the germ-plasm, has both theoretical and actual qualities different from it, and any changes in these must inevitably be communicated, by the action of hormones or other physiological mechanisms.

A brief paraphrase of the foregoing discussion may be useful in emphasizing some of the more important matters which have been touched upon. It is readily apparent that the assumption of the inheritance of acquired characters, after a long period of tolerance, with but little research activity bearing upon its principal claims, is coming in for a large share of attention from the experimentalist, and there seems a fair prospect that decisive facts may be obtained within a period, very brief in comparison with the century since the principal tenets of the theory were first formulated. Already results are available which have been obtained by cultures of animals from paramoecia to mammals, and of plants from bacteria to the higher seed-plants.

A critical consideration of the available information seems to justify the following generalizations:

External agencies acting upon bacteria, crustaceans, beetles, fungi, and some of the higher types of seed plants have been seen to result in the appearance of new types or genotypes, which have been found to transmit their characters perfectly through so many generations as to indicate practical permanency.

In the greater majority of such cases of changes in heredity, inclusive of Tower's cultures of beetles, Woltereck's experiments with *Daphnia,* Morgan's results with flies, and my own ovarial treatments of seed plants the germ-plasm was exposed to the excitation of unusual climatic factors, irradiation, concentrated nutritive media, or of solutions of sugar or inorganic salts.

The new qualities were seen to be fully displayed, and to appear in a mutational manner in all of these instances, although the new head form acquired by *Daphnia* in Woltereck's experiments did not become fixed and fully heritable until the organism had been kept under the influence of the exciting agency for an extended period, nearly two years. The most recent and one of the most interesting series of results are those which show that the influence of environic factors upon hybridizations by excitation of the germ-plasm may alter materially the results of the unions of identical pairs. This seems to have been first suggested by De Vries and to have been seen by MacDougal in hybrids of mutants of *Œnothera,* while it has been established beyond doubt by the extensive and conclusive results of Tower in crossing beetles under various conditions that environic agencies may exert a very marked effect upon the dominancy of paired characters and the general composition of hybrid progenies. A different phase of the matter is represented by the experiments of Kammerer, in which, characters constituting temporary sexual dimorphism mendelize when paired. Aberrants, sports or mutants have been seen to arise and perpetuate themselves under unusual culture conditions in yeasts and bacteria, their survival being dependent upon pedigreed cultures in some cases; and the successive generations were those resulting from fissions, although in some cases spores were interposed.

Many of the purely accommodative adjustments displayed by these organisms and by *Paramœcia* as well as the extremes of variability induced by external agencies and continued by selection, do not become fixed and are not transmissible even in a series of generations by fission. The recent work of Pringsheim, however, shows that some alterations in the way of accommodations or functional responses of yeasts and bacteria to unusual temperatures, culture media, and toxic substances become fixed and transmissible both by fission and through spores. It is not clear, however, that the differential action of the exciting agent upon soma and germ can be made out, and perhaps nothing more definite might be said than that both are directly and simultaneously exposed and exhibit coincident reaction.

When we pass to a consideration of the results of Zederbauer and Klebs, however, the evidence becomes much more decisive. A *Capsella* was found growing at an elevation of 2,000 to 2,400 meters in Asia Minor which had hairy stems, 2–4 cm. long xerophytic leaves, and reddish flowers. This plant had been evidently introduced from the lowlands by man along a route that has been in use for more than 2,000 years. The *Capsella* of the lower plains forms a stem 30–40 cm. high, has whitish flowers and broad leaves; when its seeds are taken to elevations with climates comparable to the above, individuals are developed duplicating those of the highlands, so that the characteristic features of this alpine form are clearly direct somatic reactions; and that they have become fixed and fully transmissible is demonstrated by the fact that in a series of generations grown at lower levels the stem characters, as well as those of the reproductive branches and floral organs, retained their alpine acquired characters, although the leaves, as might be expected, returned to a mesophytic form with broad laminæ.

The results obtained by Klebs include divergences of stem habit, number and structure of floral organs in *Sempervivum* which are not capable of being interpreted as functional or adaptive responses to the agencies which called them out and were found to be fully transmissible

by seeds, in which case it is fairly clear that somatically
produced characters have been impressed upon the germ-
plasm and carried by it to succeeding generations. The
structural and functional features displayed by *Semper-
vivum* in these laboratory experiments are not adaptive
in any sense in contrast with those of *Capsella,* which are
direct responses.

The actual transplantation of organisms from one lo-
cality to another, as a method of experimentation, prom-
ises the results of highest value and widest significance,
especially when taken in connection with analytical lab-
oratory cultures. This method of approach is one which
may yield evidence of the greatest value upon the influ-
ence of isolation and other geographical factors, but is
also one which allows the repetitive or mnemonic effects
to be evaluated. When supplemented by laboratory
analyses and cultures to determine the nature of altera-
tions induced, such methods promise results of the great-
est value. A series of plantations including locations from
mountain tops to the seashore has been established in con-
nection with the Desert Laboratory in accordance with this
idea, and in addition to the interchange of species from
the various localities a number of introductions have
been made from eastern America. Negative or positive
results of sufficient inclusiveness to permit analyses as to
the nature of the exciting agency and the permanence of
the response are yet available.

Some of the characters called out by environic agencies
may be retracements, or regressions, as the reappearance
of spines in cacti, or they may be awakened latencies or
organizations *de novo.* Some of the responses may result
in sexual dimorphism, while in others the induced char-
acters may be sex-limited. The alterations induced by
external agencies may be cumulative or mutative as to
appearance or organization, and they may be permanent
upon first appearance, or on the other hand may need
generations of repetition before becoming fixed. And
lastly the changes may be orthogenetic or heterogenetic
as to direction, adaptive and accommodative or correla-
tive, or wholly inutile as to their functional relations.

THE NATURE OF GRAFT–HYBRIDS

PROFESSOR DOUGLAS HOUGHTON CAMPBELL

STANFORD UNIVERSITY

THE possibility of hybrids arising as the result of grafting long has been a mooted point and the subject has given rise to much discussion.

The history of the small number of graft-hybrids that have hitherto been recorded is small and is not as complete as might be wished; indeed it has been claimed repeatedly that these supposed graft-hybrids are not really such but have been produced by the ordinary method of cross-fertilization. The most famous of these graft-hybrids is the much discussed *Cytisus Adami* which originated at Vitry near Paris about 1826. This was said to have been the result of grafting *Cytisus purpureus* upon *C. laburnum*. A series of supposed graft-hybrids is also recorded resulting from grafts between a thorn, *Cratægus monogyna,* and the medlar, *Mespilus germanicus.* Three of these graft-hybrids were secured by Bronvaux. The hybrids in this case were not all alike and were given special names and the genus *Cratægomespilus* was proposed for these bi-generic hybrids.

Of the recent opponents of the graft-hybrid theory the best known is the distinguished botanist Professor E. Strasburger, of Bonn. Strasburger made a careful cytological study of *Cytisus Adami* which has been retained in cultivation ever since its origin some eighty-five years ago. Strasburger came to the conclusion that *Cytisus Adami* was a real sexual hybrid and not a graft hybrid. He believes that if the latter were true the nuclei of the hybrid would show a double number of chromosomes. This, of course, implies that in hybrids arising otherwise than sexually, assuming that a nuclear fusion would precede the formation of such a hybrid, there would be no

41

reduction division of the nuclei comparable to that which normally occurs before the fusion of the sexual cells in normal fertilization.
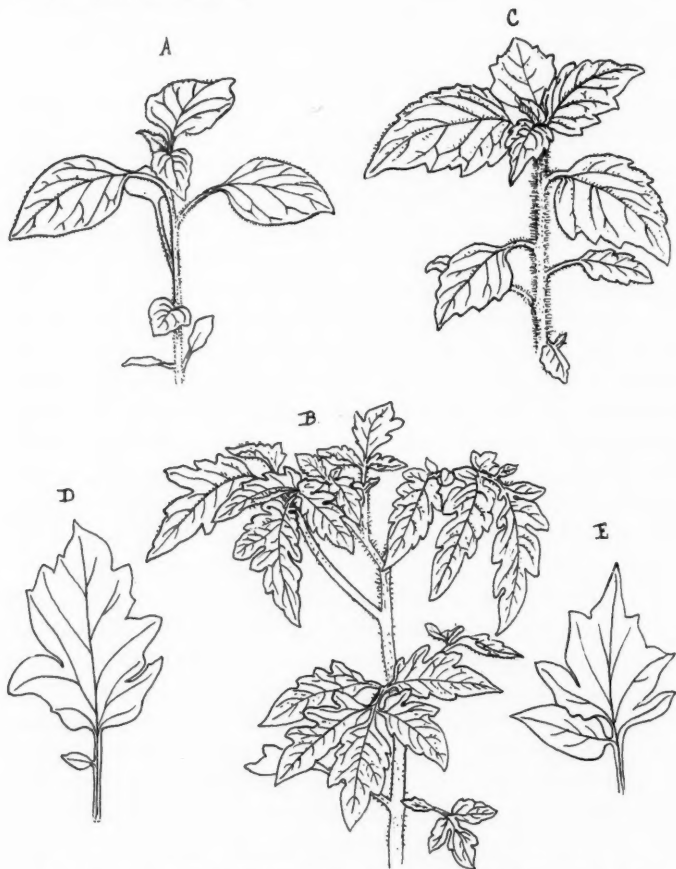

Fig. 1.   *A*, seedling of the black nightshade, *Solanum nigrum*; *B*, seedling of a tomato, *S. lycopersicum*; *C*, shoot of the graft-hybrid, *S. tubingense*; *D, E*, leaves of the graft-hybrid, *S. proteus*.   (All figures after Winkler.)

Němec,[1] however, believes that a reduction division does occur, and there is, therefore, no reason to expect

[1] Němec, B., ''Zur Mikrochemie der Chromosomen,'' *Ber. der deutsch. Botan. Gesellsch.*, 27, 46, 1909.

an increase in the number of chromosomes in the cells of the hybrid. If such a reduction does occur *Cytisus Adami* would show the same number of chromosomes as *C. laburnum* which has the same number as *C. purpureus*.

The study of graft-hybrids has assumed a new interest through the very important recent investigations of Professor H. Winkler, of Tübingen. These investigations prove beyond question that graft hybrids are possible, and the numerous experiments carried out with every possible precaution and showing remarkable ingenuity as well, furnish by far the most important study on the nature and origin of graft-hybrids that has yet been published. These experiments are being further developed by Professor Winkler but the results already obtained are of the greatest interest and value.[2]

The fact that hybrids may arise as a result of grafting touches some of the fundamental problems of heredity, and this makes these papers of Professor Winkler of the highest importance to all students of heredity, and they deserve much wider attention than they have as yet received.

The plants chosen by Winkler for his experiments were the black nightshade, *Solanum nigrum*, and two varieties of the tomato, *Solanum lycopersicum*. These two species are very distinct, and indeed many botanists regard the tomato as belonging to a distinct genus *Lycopersicum*, so that Winkler's graft-hybrids might be regarded as bigeneric like the *Cratægo-mespilus* graft-hybrids.

The methods by which Winkler secured his graft-hybrids were extremely ingenious. Seedlings of the night-

[2] 1. "Uber Propfbastarde und pflanzliche Chimæren," *Ber. d. deutsch. botan. Gesellsch.*, 25, 568–576, 1907.

2. "Solanum tubingense, ein echter Propfbastard zwischen Tomate und Nachtschatten," *ibid.*, 27, 595–608, 1908.

3. "Weitere Untersuchungen über Propfbastarde," *Zeitschr. für Botanik*, 1, 315–345, 1909.

4. "Uber die Nachkommenschaft der Solanum-Propfbastarde und die Chromosomenzahlen ihrer Keimzellen," *ibid.*, 2, 1–38, 1909.

5. "Uber das Wesen der Propfbastarde (Vorläufige Mittheilung)." *Ber. der deutsch. botan. Gesellsch.*, 28, 116–118, 1910.

shade and of the tomato were decapitated and reciprocal grafts were made. In making these unions the graft was cut either wedge-shaped or saddle-shaped at the point of junction with the stock. The graft and stock united readily whether the nightshade or tomato was used as the stock. After the union was complete the plant was again decapitated, the cut being made through the region where the union had taken place. The cut surface thus exposed is composed of tissue derived from the two members of the union and from this cut surface a callus soon develops from which numerous adventitious buds quickly arise. It was thought that from some of these adventitious buds arising at the point of the junction of the graft and stock there might be produced shoots which would combine the characteristics of the two, or at least might be composed of tissue derived from the two parents.

Naturally the great majority of the shoots arising from the cut surface of the stem were either pure nightshade or pure tomato. But finally shoots were observed which were evidently of mixed origin. The first of these graft-hybrids were obviously composed of pure elements derived from the two parents. Some of these shoots were almost equally divided by a median line on one side of which the organs—stem, leaf, etc.—were those of the nightshade, while on the other the organs were evidently derived from the tomato. Sometimes a leaf was nearly equally divided. In most cases one or the other of the parents predominated, but there was no intermediate region between the two kinds of tissues and organs. It is clear that such monstrous forms, for which Winkler proposes the name "chimæra," are not hybrids in any true sense of the word, but have arisen from buds in which there was a mere mechanical coalescence of tissue from the two parent forms at the junction of the stock and graft.

Further experiments, however, resulted in the production of shoots in which the characteristics of the two parents were so intimately combined, that their discov-

erer felt warranted in assuming that these were really
hybrids, probably arising from the actual fusion of cells
derived respectively from the nightshade and the tomato,
this fusion taking place where the graft had united with
the stock.   This cell-fusion was assumed to involve a
fusion of the nuclei as well, analogous to the fusion of the
egg-nucleus with the generative nucleus of the pollen
tube in normal fertilization.

Several types of these graft-hybrids were produced
and to these specific names were given.

The first genuine graft-hybrid was called *Solanum
tubingense* and it has since been produced several times
and has been propagated by cuttings and distributed to
various botanical gardens.   During the past summer I
had an opportunity of seeing this graft-hybrid growing
well in the botanical gardens of the University of Munich.

Solanum tubingense is intermediate in external appear-
ance between the nightshade and tomato but is rather
nearer the former (see Fig. 1, *C*).   The nightshade (*A*)
has simple, smooth-edged, oval leaves and an almost
smooth stem.   The tomato (*B*) has compound leaves with
sharply serrate leaflets and all of the varieties are
strongly hairy.   The hybrid (see Fig. 1, *C*) has simple
leaves but they are sharply serrate or often slightly lobed
like the leaflets of the tomato, and both stem and leaves
are abundantly provided with hairs.

The flower in *Solanum tubingense* is also intermediate
in character.   The nightshade has small white flowers
with a smooth calyx whose lobes are very short.   The
flower of the tomato is much larger, bright yellow in
color and the lobes of the calyx are hairy and very much
longer than those in the nightshade.   The hybrid has
flowers which are intermediate in character.   They are
larger than those of the nightshade but much smaller
than those of the tomato, but like the latter the flowers
are a pronounced yellow.   The calyx lobes are two or
three times as long as those of the nightshade but much
shorter than those of the tomato.   Like the latter, how-

ever, there are numerous hairs upon the calyx lobes which in the nightshade are almost smooth.

The fruit of *Solanum tubingense* is very much like that of the nightshade but is rather larger, and although it is black in color there are some traces of the red or yellow color of the tomato.

Four other well-marked graft-hybrids were secured to which were given the names *Solanum proteus, Solanum darwinianum, Solanum koelreuterianum* and *Solanum gaertnerianum.*

The first of these originated in a most peculiar fashion. A chimæra was obtained which consisted of *two* hybrid components. One of these was the before mentioned *S. tubingense* while the other was a hybrid which was more like the tomato. This chimæra soon divided into two branches one of which was pure *S. tubingense* and the other the new hybrid, *S. proteus.* The latter was then removed and rooted and further propagated by cuttings. This species has very variable leaves (see Fig. 1, *D, E*) which on the whole are more divided than those of *S. tubingense,* while in the characters of the flower and the fruit it is more like the tomato than like the nightshade.

Both of the forms *S. koelreuterianum* and *S. gaertnerianum* were produced more than once and they are respectively more like the tomato and nightshade but each differs in important particulars from either of the parents.

The form, however, which is of the greatest interest is the hybrid to which Winkler gave the name *S. darwinianum,* the third to result from his experiments. This hybrid arose in a quite different manner from the others and great ingenuity was shown in isolating and propagating it. The shoot from which this hybrid originated was a chimæra which developed from a graft of a tomato upon a nightshade. This chimæra was made up principally of pure *Solanum nigrum,* but a small portion of it consisted of tissue which was different from any

of the forms which had yet been discovered. The chimæra instead of being made up of two portions united longitudinally was composed mainly of tissue evidently of pure *Solanum nigrum* origin. A small strip, however, near its base was of a different character. This region consisted of a single leaf, and a small amount of tissue lying below belonging to the stem. The same form was secured a second time where it developed from a five-fold chimæra derived from *S. proteus*. Unfortunately, it was not possible to propagate this second specimen.

In order to isolate this new form it was necessary to cause the axillary bud belonging to the single leaf to develop into a shoot. This was finally successful after four decapitations of the *Solanum nigrum* shoot above it. The final result was a branch which was very different from any of the previously developed forms and it was named *Solanum darwinianum*. The point of special interest in connection with this form is that of all graft-hybrids which Winkler secured, this seems to be the only one which is likely to prove a hybrid in the strict sense of the word. This point, however, will be brought out later in the discussion of the real nature of these graft-hybrids.

All of the hybrids were propagated further by cuttings and with the single exception of *Solanum koelreuterianum,* were made to produce ripe fruit which in all cases was more or less intermediate in character between the fruit of the nightshade and the tomato. In *Solanum darwinianum,* however, the fruit was all sterile and no perfect seeds were formed. The fruit itself is a small round berry like the fruit of the nightshade in form, but having the color and structure of the tomato. In *Solanum koelreuterianum* the young fruit set but failed to reach maturity.

Of the hybrids *Solanum tubingense* is the most fertile and produces fruit very abundantly. A considerable number of the fruits, however, are sterile or "parthenocarpic" and the seeds in no cases reach their full de-

velopment. Nevertheless Winkler was able to make
these seeds germinate and the second generation of the
plants was reared. *S. gaertnerianum* produces fruit only
in small numbers but the seeds are perfectly developed
and germinate readily, the same being true of *S. proteus*.

<center>REVERSIONS</center>

Winkler observed a number of cases where the graft-
hybrid reverted to one or other of the parent forms.
Similar cases of reversions have been recorded for
Cytisus Adami and *Cratægo-mespilus*. These rever-
sions were studied with special care in his first hybrid
S. tubingense. In several instances where the plant was
cut off below the first lateral bud numerous adventitious
shoots arose from the cut surface, and while some of
these were pure *S. tubingense,* others were pure *Solanum
nigrum,* the parent species which is nearer to *S. tubin-
gense*. In a similar manner *S. proteus* was observed
frequently to revert to the tomato, but in no case was
there reversion to the nightshade.

Sometimes spontaneous reversions occur. Thus in *S.
tubingense* the apex of a plant was noted which had sud-
denly assumed the characters of *S. nigrum*. Winkler
gives an excellent photograph of this plant. In other
cases shoots of mixed nature were seen, some having the
structure of chimæras, half nightshade and half the
hybrid form. In these mixed shoots the inflorescence
had flowers of two sorts belonging respectively to the
nightshade and to the hybrid. Similar mixed inflores-
cences have also been observed in *Cytisus Adami*.

<center>*The Second Generation*</center>

In *S. proteus* and *S. gaertnerianum* perfect seed is de-
veloped and germinates readily. *S. tubingense* which
sets fruit freely never has the seed fully developed but
as we have already stated Winkler succeeded in germi-
nating these seeds and rearing plants from them. He

explains the failure of the seeds to develop fully to the fact that the fruit of the hybrid, which closely resembles that of the nightshade, ripens before the seeds have had time to complete their development. The tomato fruit requires a very much longer time for maturing than does the berry of the nightshade and a correspondingly longer time is needed for the seed to be perfected; and he thinks that the longer time required for the seed development in *S. tubingense* is an inheritance from the tomato parent, while the fruit is mainly of nightshade derivation.

All of the seedlings derived from these hybrids reverted absolutely to that parent form which the hybrid more nearly resembles. Thus the seedlings of *S. tubingense* and *S. gaertnerianum* are pure *S. nigrum,* those of *S. proteus* pure tomato. This behavior also corresponds to that of the very few cases where seedlings have been secured from *Cytisus Adami,* these in all cases proving to be pure *Cytisus laburnum.*

Of the *Cratægus-mespilus* hybrids only one, *Cratægo-mespilus asnièresi* produced seed capable of germinating. These seedlings were not reared to maturity but so far as could be judged from the young plants, were pure *Cratægus monogyna,* the parent which the hybrid more nearly resembled.[3]

The third and fourth generations of the *S. tubingense* seedlings retain perfectly the characters of *S. nigrum* and the same is the case when they are cross-pollinated by *S. nigrum.* Attempts to cross *S. tubingense* with the tomato resulted in the formation of fruit but no seeds were developed. It may be also recorded that crosses between the two parent forms, the nightshade and the tomato, were without any result.

S. proteus crossed with the two parent forms produced seed when crossed with the tomato to which it stands the nearer, and sterile fruit when crossed with *S. nigrum.*

[3] Noll, F., "Die Propfbastarde von Bronvaux," *Sitzungsber. der niederrheinische Gesellsch. für Natur- und Heilkunde,* 1905.

As yet no seed has been obtained from crosses between the graft-hybrids themselves.

The Nature of Graft-Hybrids

Winkler concluded at first that all the graft hybrids except the chimæras probably arose from actual cell fusion and might be compared directly with hybrids arising from true fertilizations. It was suggested by another student of graft-hybrids, Bauer,[4] that these apparent true hybrids might also be chimæras of a type which he has called "periclinal," *i. e.*, the outer tissues are derived from one parent, and the inner tissues from the other, but none of the tissues themselves are of hybrid nature. This hypothesis seemed the more probable from the results of investigations of MacFarlane upon *Cytisus Adami*[5] in which he showed that the epidermal tissues were strikingly like those of *C. purpureus* while the inner tissues were like those of *C. laburnum*. An investigation of the *Cratægo-mespilus* hybrids revealed a similar state of affairs.

Acting on this suggestion Winkler made a careful cytological study of his hybrids and found that four of them were indeed periclinal chimæras. But one of them seemed to be a real hybrid resulting apparently from a fusion of cells at the junction of the graft and stock.

The nuclei in the nightshade and the tomato differ very much in the number of the chromosomes so that the determination of the origin of the tissues in the hybrid is made comparatively easy. The chromosome number in the sporophytic tissue is twenty-four in the tomato and seventy-two in the nightshade. These numbers were found in the tissues of all of the graft hybrids except *S. darwinianum* where the reduced number of the

---

[4] Bauer, Erwin, "Propfbastarde," *Biologisches Centralblatt*, 33, No. 15, 497–514, 1910.

[5] MacFarlane, J. M., "A Comparison of the Minute Structures of Plant Hybrids with those of their Parents, and its Bearing on Biological Problems," *Trans. Roy. Soc. of Edinburgh*, 37, 203–286, 1895.

chromosomes in the germ cells was found to be twenty-four, which was to be expected if these were derived from cells with forty-eight chromosomes; *i. e.*, one-half the number of the twenty-four plus seventy-two chromosomes of the two parents. It is assumed by Winkler that a reduction in the number of chromosomes follows the fusion of the cells. He says:

> This chromosome number, *i. e.*, forty-eight, is most readily explained by the assumption that in the formation of the graft hybrid a nightshade cell (with seventy-two chromosomes in its nucleus) and a tomato cell (with twenty-four chromosomes) united. The resulting cell, from which the subepidermal layer at the apex of the darwinianum hybrid arose, had a nucleus with ninety-six chromosomes which then underwent a reduction division resulting in forty-eight chromosomes.

This study of the tissues of *S. darwinianum* indicates that the subepidermal tissue from which the sporogenous cells develop is of genuine hybrid nature arising from a fusion of cells including the nuclei derived from the two parent forms.

In his latest paper (5) Winkler gives a brief summary of his conclusions which are as follows:

> Hybrids may be arranged in two groups, sexual and graft hybrids. The latter may be divided into three classes according to the theoretical possibility of their method of origin, viz.: (1) Fusion graft-hybrids arising from a fusion of two somatic cells derived from distinct species. (2) "Influenced" ("Beeinflussungs Propfbastarde") graft-hybrids which arise from specific influences of one graft component upon the other without cell fusion (as through chemical substances, translocation of cytoplasm, etc.). (3) Chimæras, in which specifically pure cells from both graft components are combined to form a new individual. These chimæras may be: (*a*) Sectorial chimæras in which the two sorts of cells in the growing point are divided by a longitudinal plane. (*b*) Periclinal chimæras in which the periclinal cell layers of the growing point are furnished respectively from one or the other parent form. (*c*) Hyperchimæras in which the growing point is made up of a mosaic of cells derived from the two parent forms.

The first of Winkler's graft-hybrids were unmistakably chimæras of the first type. Of his later graft-hybrids to which he gave special names, all except *S.*

darwinianum are periclinal chimæras. This is true also of *Cytisus Adami* and the *Cratægo-mespilus* hybrids. Thus *S. tubingense* has its epidermal region derived from the tomato while the inner tissues including those which give rise to the sporogenous cells are of night-shade origin. In *S. proteus* the reverse is the case. This explains all cases of reversion to the parent forms and also the character of the seedlings which in the one case are pure nightshade and in the other tomato, this being due to the fact that the spores (pollen spores and embryo-sacs) arise from sub-epidermal tissues derived from the nightshade or the tomato as the case may be.

These remarkable experiments of Winkler's must be of the greatest interest to all students of the problems of heredity. They emphasize a fact, too often over-looked, that it is not always safe to apply to the study of plants the data of zoology. It must be remembered that in the evolution of the higher plants there has been a constant tendency toward a reduction of the sexual re-productive parts. Many biologists quite ignore the fact that the flowering plant, as it is generally understood, is a purely sexless organism. The so-called sex organs, stamens and carpels, are not such at all, but are non-sexual sporophylls.

The sexual generation of the highest seed plant is a far simpler organism than that of the moss or fern and the sex organs are correspondingly simpler. Moreover the development of the sex cells and the extraordinary correspondences in nuclear structure, the reduction divisions and the mechanics of fertilization must have developed quite independently of these phenomena in animal cells, since the two great divisions of organisms, plants and animals, parted company for good long be-fore the elaborate structures found in the higher mem-bers of the two series were developed. Hence it by no means follows that what is true in one case must neces-sarily follow in the other.

With the subordination of the sexual generation of

the higher plants there goes a high degree of regenerative power, a great contrast to the very limited capacity for regeneration shown by the highly organized animals where new individuals can only arise through sexual reproduction. This great power of regeneration in plants is accompanied by much less specialized cells and a very imperfectly marked individuality of the organism as a whole. Any seed plant may be regarded as a colony of individuals since the parts are repeated indefinitely and can be made to regenerate the whole plant. The power of regeneration shown by almost any part of the plant, even a single cell in some cases, renders any theory of a special germ plasm out of the question in the case of plants, however plausible such a theory may appear when applied to animals.

It is not then so very extraordinary that this regeneration of the plant from somatic cells should be carried so far as to involve cell fusions such as Winkler believes preceded the formation of his *Solanum darwinianum.* Even if this should not be proved, his experiments show beyond question the existence of graft-hybrids of a sort quite inconceivable in any animals except very low types, such as corals where it is by no means impossible that similar graft-hybrids might be developed.

It is this positive demonstration of the reality of ''vegetative'' or ''somatic'' hybrids which gives the experiments of Winkler their greatest value, and it is to be hoped that they will serve as a stimulus to other work in the same direction which may well have a great influence upon the drift of biological speculations dealing with the laws of heredity.

A DOUBLE HEN'S EGG[1]

DR. J. THOMAS PATTERSON,

UNIVERSITY OF TEXAS

DOUBLE hens' eggs have always attracted much attention and the literature covering the records and descriptions of the various kinds has become extensive. In a comparatively recent paper published in this journal, Parker[2] has reviewed the main contributions to the subject and from a consideration of these, together with his own observations on five double eggs, has been led to the conclusion that at least two factors are involved in the production of such eggs. According to him, double-yolk eggs are produced when there is a simultaneous discharge of two yolks from the ovary, whether these are derived from a single follicle or from two separate follicles; while inclosed eggs may be the product of a normal ovary and probably are produced through the abnormal action of the oviduct, "in that a yolk normally supplied by the ovary may be abnormally covered, retained and inclosed in another egg." Parker therefore classifies the factors concerned in the formation of double eggs as ovarian and oviducal. Three types of double eggs result from the action of these two factors: (1) those in which the yolks have been derived from an abnormal ovary but have traversed a normal oviduct, (2) those produced by an ovary and an oviduct both of which have functioned abnormally, (3) and those in which the yolks have come from a normal ovary but have passed through an abnormal oviduct.

The egg described in the following pages clearly belongs to the third type, and since it possesses certain

---

[1] Contributions from the Zoölogical Laboratory of the University of Texas, No. 107.

[2] "Double Hens' Eggs," by G. H. Parker, AMERICAN NATURALIST, Vol. XL, 1906.

FIG. 1. A photograph of the egg. $\times \frac{2}{3}$.

FIG. 2. In this photograph the egg is shown after an incision had been made in the outer membrane and the upper margin of the cut turned back so as to reveal the inner membrane. Note that while the outer membrane shows deposits of lime, especially on the smaller end of the egg, the inner membrane, on the contrary, is entirely free from such deposits. $\times \frac{2}{3}$.

FIG. 3. This shows both membranes cut open and the contents of the egg clearly revealed. The dark mass lying just to the left of the inclosed egg is the yolk of the inclosing egg. $\times \frac{1}{2}$.

peculiarities that have not appeared in any previous account, seems to be worthy of record. It should be kept in mind, however, that the value of such descriptions does not come from the morphological features revealed, no matter how bizarre these may be, but from the fact that considerable light is often thereby shed on the complex physiology of the reproductive organs, and it is with this idea in mind that the following record is made.

The egg in question, which was kindly handed to me by Mr. S. L. Pinckney, Austin, Texas, was laid March 28, 1910. It was stated that several double eggs had been received from the flock from which this egg came, but whether they were all laid by the same hen could not be ascertained. The egg was large, measuring 85 mm. in its long axis and 62 mm. in the short axis, and was slightly smaller at one end than at the other, so that we may speak of the blunt and pointed ends. It was practically a soft-shelled egg, in that the amount of lime deposited on the shell-membrane was very small, and for the most part was collected into little nodules scattered about over the surface (Figs. 1 and 2). A microscopical examination of the shell-membrane did not reveal anything unusual, for it consisted of the two characteristic layers, a thick outer and a thinner inner; but on cutting it open I was surprised to find another shell-membrane lying almost directly beneath it (Fig. 2). The two membranes were separated by the very thinnest layer of watery albumen. This second or inner membrane was in every way normal, and perfectly white, but was entirely void of lime deposits, reminding one very much in its general appearance of the membranes on eggs which have just reached the isthmus.

The contents of the inner shell-membrane consisted of much albumen in which were imbedded a hard-shelled egg and a yolk (Fig. 3). Upon examination the inclosed egg was found to be perfectly normal in every respect, and its yolk contained a healthy blastoderm. The inclosed yolk, although normal in structure, was much dis-

torted, owing to the pressure exerted upon it by the approximation of the hard-shelled egg. The albumen closely adhered both to the egg and to the yolk, but much of it was of a liquid nature, as was indicated by the ease with which it flowed out of the cut first made in the inner membrane.

The accompanying diagram will make clear the relation of the various parts of this interesting monstrosity (Fig. 4). The inclosed egg lies toward the pointed end
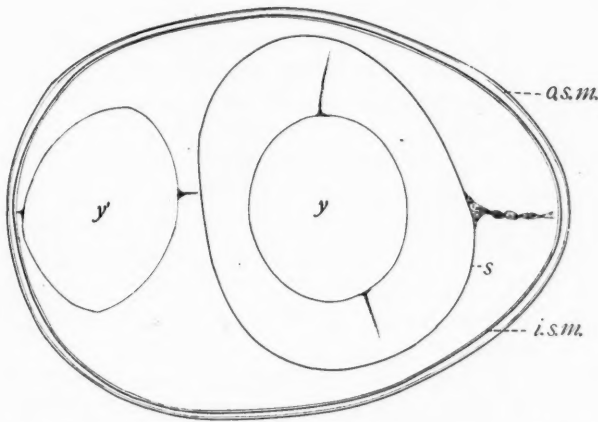

FIG. 4. Diagram of a median section of the egg. *i.s.m.*, inner shell-membrane, the two lines representing its two layers; *o.s.m.*, outer shell-membrane; *s*, shell of the inclosed egg; *y*, yolk of the inclosed egg; *y'*, yolk of the inclosing egg. Natural size.

of the inclosing egg, and its long axis meets the corresponding one of the double egg at an oblique angle. On account of this inclination of the inclosed egg its pointed end lies nearer to the blunt than to the pointed end of the inclosing egg. The inclosed yolk occupies the blunt end of the inclosing egg and is considerably distorted by pressure. The chalazæ are but poorly developed, but the axis formed by a line passing through their points of attachment to the vitelline membrane approximately coincides with the long axis of the inclosing egg, showing that the yolk has maintained its original orientation.

As I have pointed out above, the most interesting question regarding this egg pertains to the physiology of its formation. Parker states that two hypotheses have been advanced to explain how inclosed double eggs are formed. According to one of these, which was first advocated by Panum,[3] the inclosed egg remains in the distal part of the oviduct until overtaken by a second one, when both are then surrounded by a common envelope; according to the other a completely formed egg is carried by antiperistalsis back up the oviduct, where it meets a second one, and the two passing down become covered by a second shell and are laid. It seems quite evident from the description of the egg just given that it is the product of antiperistalsis, but the especial interest lies in the fact that this process has taken place twice.

The first antiperistalsis took place immediately after the hard-shelled egg was formed, and of course caused its migration to the upper or proximal end of the oviduct where it met the second egg. This meeting must have taken place very close to the infundibulum, for otherwise the yolk of the second egg would have possessed much larger chalazæ.

The second antiperistalsis occurred immediately after the inner of the two shell-membranes had been laid down, and must have succeeded in carrying the double egg up the oviduct to a point where albumen is secreted, that is, to a place slightly above the beginning point of the isthmus; for it is only on this assumption that we are able to explain how a thin layer of albumen came to exist between the two shell-membranes. The small amount of lime deposited on the outer of the two shell-membranes indicates that the egg did not remain long in the uterus, but must have been laid shortly after having entered that organ.

In many respects this egg conforms to the facts already seen in the inclosed types of double eggs; thus the in-

[3] Untersuchungen über die Entstehung der Missbildungen zunächst in den Eiern der Vögel,'' by P. L. Panum, Berlin, 1860.

closed egg lies near the pointed end of the inclosing one, and it was laid during the time of year when such eggs most frequently appear, that is, in the winter or spring; but it differed in one rather important respect. The pointed end of the inclosed egg does not lie in the same direction as that of the inclosing one. This unusual position of the inclosed egg doubtless has been brought about by crowding, and does not indicate necessarily that it was at first incorrectly oriented.

Among the more important things so far revealed by a study of inclosed double eggs is the light thrown on the problem of the orientation of the egg in the oviduct, a problem in which the writer has been deeply interested. These eggs clearly demonstrate that when an egg has once entered the oviduct its original orientation in that organ is maintained during the formation of the envelopes, no matter to what extent it may have been moved up and down the reproductive passage. This fact strongly supports the conclusion reached by the writer[4] in a recent contribution, in which it was pointed out that the definite orientation of the egg in the reproductive passage is not a matter of chance, but is something that is handed on to the oviduct by the ovary, that is to say that the ova in the ovary have a definite polarity which is passed on to the oviduct through the mechanism of the infundibulum.

[4] "The Early Development of the Hen's Egg, I., History of the Early Cleavage and of the Accessory Cleavage," by J. Thomas Patterson, *Journal of Morphology*, Vol. 21, 1910.

NOTES AND LITERATURE

HEREDITY

One of the most important papers relating to heredity that has appeared in recent months is that of Tower, dealing with hybridization investigations with species of the genus *Leptinotarsa*.[15] I shall not here attempt an extensive review of this paper, but mention it rather as a means of calling attention to its importance and suggesting that any one interested in theoretical discussions of heredity should not fail to read it. Tower has done an immense amount of work with this genus. His results lead him to accept the factorial hypothesis as an explanation of Mendelian phenomena but to discard wholly the de Vriesian interpretation of these factors. The most important contribution in this paper is the apparent fact that the environment at the time when eggs are fertilized may change very materially the nature of the hereditary factors. It is unfortunate that the author does not give more details in connection with this conclusion. The data he does give are mixed and contradictory. There has possibly been an error in printing Tower's paper, but if not there was a serious error in its preparation, as will be seen from the following. In experiment 409, which was several times repeated, the results were exactly as if one of the parents had been a heterozygote between the two species. F_1 consisted of two types, one of which was identical with the female parent and the other intermediate between the two. The one like the female parent bred true to that type, while the other behaved in all respects as a heterozygote. In experiment 410 the same two species were utilized, but the temperature and humidity conditions at the time the eggs were fertilized were made quite different. This experiment, which was repeated eleven times, gave in every case ordinary Mendelian phenomena.

In experiments 409/411, which was performed seven times, one set of eggs from the same cross as above was produced under conditions identical with those of experiment 409, and in

[15] Tower, Wm. L., "The Determination of Dominance and the Modification of Behavior in Alternative (Mendelian) Inheritance, by Conditions Surrounding or Incident upon the Germ Cells at Fertilization," *Biol. Bull.*, XVIII, No. 6, May, 1910.

each case gave identical results with those of 409; that is, F_1 consisted of two types, one heterozygote, and the other homozygote of the maternal type. Using the same individuals which produced a set of eggs of this kind to secure another set of eggs produced under the conditions of experiment 410, the results in each of the seven experiments gave F, which behaved in all respects as a homozygote of the maternal type. This fact is set forth in considerable detail and Plate III illustrates it just as here described. This result occurred in all cases whether the set of eggs produced under the conditions of experiment 410 was produced before or after the set which gave the results of experiment 409. Now the remarkable thing about this experiment is this. While in experiment 410 the results in each of the eleven cases gave ordinary Mendelian heterozygotes in F_1, in each of the seven cases of 409/411 the eggs produced under the same conditions as 410 gave homozygotes of the maternal type. Thus, the conditions of experiment 410 in eleven cases gave one result, in seven other cases they gave an entirely different result, and the only difference in the conditions was that in 409/411 the female either had produced or was in the future to produce a set of eggs under the conditions of experiment 409 (p. 295).

Thus, on page 294, in describing experiment 410, it is stated that experiment 410 gave F_1 all heterozygote; on page 330 it is stated that experiment 410 gave F_1 all homozygote of the maternal type; on page 295–6, in describing experiment 409/411, it is stated that, under the conditions of experiment 410, this experiment gave F_1 all homozygotes of the maternal type; and on page 304 it is stated that experiment 409/411, when performed under the conditions of 410, gave results identical with those of 410. These statements are directly contradictory. We must withhold judgment on this point of influencing the hereditary factors at the time of fertilizations, until Mr. Tower informs us which of these statements are correct.

In these exceptional cases, where the F_1 hybrid behaved as a homozygote of one of the paternal races, the author does not tell us whether the F_2 and later generations were each time produced under the conditions which produced the aberrant F_1. One would infer, however, that they were not, and that the change which occurred in the fertilization of the eggs which produced F_1 was permanent and not reversible. It is hoped

that he will give us fuller data on this point in future papers which are promised.

In several cases Tower mixed three species which interbred freely and left them under natural conditions for several years. A careful study of the progeny in each case showed that a new type arose, consisting of a complex of the characters of the old types, and that this new type rather rapidly replaced every other type, although some of these other types were known to be quite capable of existing under the conditions of the experiment. This would indicate that in some way the new type had a distinct advantage over the other types with which it competed for food, or possibly the repeated crossing of the types was in some way inimical to all the types except the one. Experiments of this character show that hybridization may be an important factor in the development of new varieties or possibly new species.

From the fact that when the same species are mixed together in two places where the conditions are different, the resulting type which finally wins out and becomes practically the sole representative of the mixture, is different under different conditions. Tower draws the conclusion that the conditions surrounding the germ cells at the time of fertilization "profoundly modify the behavior and the relationships of the characters entering into the crosses." This conclusion seems hardly justified. Of course it may be correct, but the well known fact that from complex crosses of this kind a great many types may result from what we know of the behavior of Mendelian characters and that these types would naturally bear different relations to the environmental conditions offers apparently a much simpler explanation of the reason for the survival of the one type under one set of conditions and another type under another set of conditions. It seems hardly necessary to assume that the conditions existing at the time of the fertilization of the egg determine the characters which were to result from the fertilization to explain this particular phenomenon.

Subsequent investigation of these new types of mixed origin showed that in all cases they occasionally produced individuals different from the general population but which in all cases exhibited characters which were present in the original parents of the complex mixture. Tower repeatedly compares this phenomenon to the phenomena which de Vries observed in *Œno-*

thera lamarckiana, and suggests that the mutations which de Vries observed are probably due to previous hybridization. This is a very interesting suggestion, but the writer is inclined to believe that the phenomena observed by de Vries were due to a different cause. It is definitely proved that in some of de Vries's mutants the chromosome numbers are different from those of the parent form. Cytological investigations have also shown that in the reduction division in these *Œnothera* mutants there are frequent irregularities in the distribution of chromosomes. It seems probable that de Vries's mutations are not the result of previous hybridizations but rather are due to irregular behavior of chromosomes in the reduction division. If this is true then the phenomena observed by de Vries would be due to a different cause from that which presumably produced the results which Tower observed. In the case of Tower's results we can explain the facts by the assumption of simple Mendelian segregation. In de Vries's work there is evidence that the phenomena are due to a different cause.

It is gratifying that Tower takes a very broad view of the factorial hypothesis of Mendelian phenomena. On page 323 he remarks:

This factorial point of view is in no wise, of necessity, to be tied to or confounded with such speculations as the id-determinant-biophore fabric of Weismann, nor with the pangene complex of de Vries, which have no foundation in fact.

This is the view which the writer has held for years and has frequently set forth in these pages. I have also frequently pointed out that we do not yet have sufficient knowledge of the physiological processes of living matter to permit us at the present time to formulate an adequate theory of the phenomena observed in hybrids. I think we can, however, point out the general nature of the causes underlying these phenomena, as I have attempted to do in my theory of Mendelian phenomena.[16] In speaking of the difference in germ cells with respect to given characters, he has the following to say:

What this difference in the gametes is we do not know, but observed behaviors are interpreted as being, most probably, due to the mechanical separation into different germ-cells of whatever it is that produces the contrasting attributes—segregation during gametogenesis.

[16] *American Breeders' Magazine,* Vol. I, No. 2.

He further remarks on page 328:

At present in biology we have no business with ultimate conceptions, and the two thus far attempted of germinal composition—the " particulate conception " and the " crystalline entity " are both equally dismal failures and equally useless as working hypotheses.

The statement on page 335, that characters which Mendelize are in the main unimportant attributes of the organism and only rarely are of importance in the struggle for existence, is a little bit strong.   Apparently it would have been better to state that those characters which have been shown to Mendelize are of this nature.   Unfortunately, most of the work of the Mendelians has been done with these superficial, easily observed characters.   I see no reason why any character whatever might not, from the failure of some chromosome to perform a usual function, give a variation which would behave in Mendelian fashion if the resulting type were capable of propagating and crossing with the parent type.

Tower's paper will undoubtedly have an important influence on biological thought, as it deserves to have.

W. J. SPILLMAN.